

DNA TESTO SINGLE PROBE TEMPERATURE VERIFICATION OF THE MASTERCYCLERS

A. SCOPE

- A.1 The Testo temperature verification system checks temperature accuracy in the block of device in the Mastercycler family to be tested. The temperature device consists of a digital display device with a temperature sensor connected. During verification, the temperatures measured are compared to specified values for 6 different block positions. The accuracy of the measuring system is +/- 0.3K for the measurement range for 35°C to 95°C. The permitted tolerance for the Mastercycler ep gradient S is +/- 0.6K (35°C) and 1.0K (95°C).

B. QUALITY CONTROL

- B.1 Protective gloves, scrubs, and a lab coat will be worn at all times when performing this procedure to prevent contamination.
- B.2 The Testo temperature verification system will be sent out for calibration on an annual basis.
- B.3 The Testo temperature verification system is safely stored and transported in the manufacturer's case provided with the instrument. Upon return from calibration, the Testo will be visually inspected for damage caused during transportation and the calibration certification will be reviewed and stored in the LAM portion of BEAST.
- B.4 The white cable attached to the single probe is fragile, handle with care.

C. SAFETY

- C.1 Protective gloves, scrubs, and a lab coat will be worn at all times when performing this procedure.
- C.2 The Mastercycler sample block may be hot immediately after the verification is complete. Handle the sensor plate with care.

D. REAGENTS, STANDARDS AND CONTROLS

N/A

E. EQUIPMENT

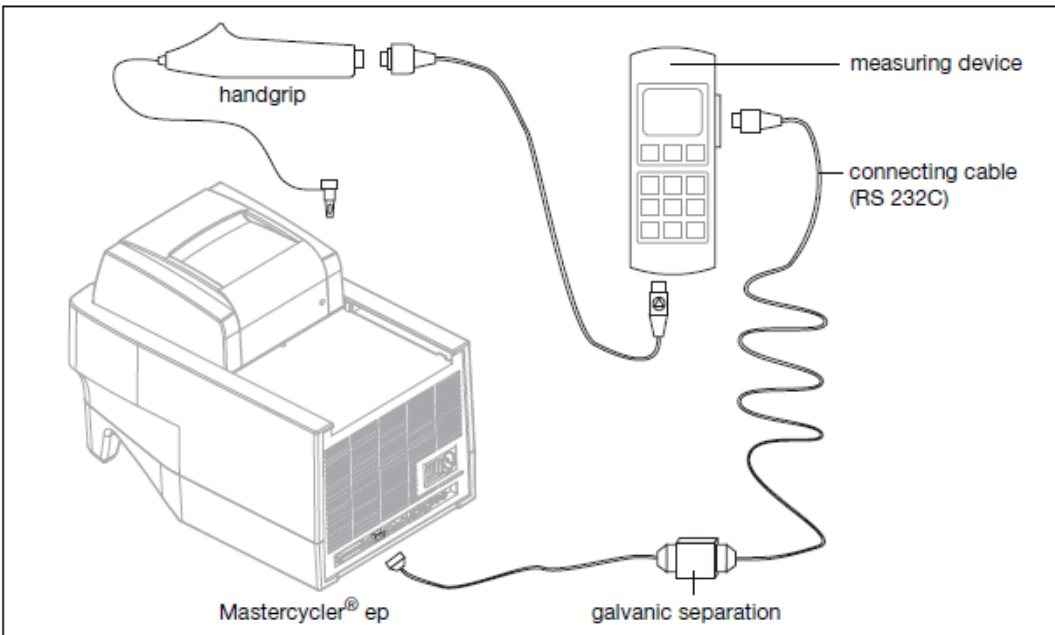
- E.1 Testo Temperature Verification System
- E.2 Mastercycler

F. PROCEDURES

- F.1 Make sure that no Mastercyclers are being utilized before starting this procedure.

Document ID	Revision	Approval	Date Published
7496	8	Supervising Criminalist - Biology	8/28/2018 9:53:16 AM

F.2 Make sure that the measuring device is switched off. Assemble the verification device by connecting the handgrip with single probe to the measuring device (Testo 950). Connect one end of the galvanic separation unit to the measuring device and the other end to the thermal cycler that is being checked. Plug the unit into the wall with the correct adapter (see diagram below).



(Diagram taken from Eppendorf Temperature Validation System Manual)

F.3 Turn on the measuring device.

F.4 On the control panel, use the arrow keys to highlight the Mastercycler undergoing temperature verification and then press "Enter".

F.5 Press the down arrow so that the newly-shown option "System" is highlighted. Press the function key labeled "Validation".

F.6 A warning message will appear on the screen informing the user to be sure they are using the appropriate temperature sensor. Press "OK".

Document ID	Revision	Approval	Date Published
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- F.7 A message will appear informing the user to connect the measuring device to the thermal cycler. Press "OK".
- F.8 Before proceeding, the lid temperature must heat up to 100°C. This step takes approximately three minutes.
- F.9 After the lid temperature has heated to 100°C, a diagram of a 96-well plate will appear on the control panel screen. A blinking red dot will indicate where to place the probe on the thermal block. Open the thermal cycler and remove the front clip. Place the temperature probe in the appropriate position and replace the front clip with the cable lead-through front clip so that the sensor cable isn't damaged. Close the thermal cycler and press "OK".
- F.10 The Mastercycler will proceed to heat up to 95°C and cool to 35°C. This process takes approximately three minutes.
- F.11 When the first run has been completed, the control panel will prompt the user to open the thermal cycler. Open the lid and press "OK".

WARNING: THE PROBE IS STILL HOT. ALLOW FOR TIME TO COOL BEFORE HANDLING THE PROBE

- F.12 Again, a diagram of a 96-well plate will appear on the control panel with a blinking red dot indicating where the probe needs to be positioned. Remove the cable lead-through front clip, reposition the probe, replace the cable lead-through front clip, close the lid, and press "OK". Repeat this step for the four remaining positions.

NOTE: A RUN CAN BE CANCELLED AT ANY TIME BY PRESSING THE "EXIT" BUTTON.

- F.13 After all six positions have been checked, a message will appear to open the thermal cycler. Press "OK". A certificate screen will appear on the control panel. Confirm that all positions tested passed. If any positions fail, repeat the verification. If any positions fail proceed to F.14. Review the verification certificate, if the failing wells are within -2°C - +1°C based on the internal validation the verification can be deemed passing. If these wells are outside this allowed range and the problem persists, perform the temperature verification using the Eppendorf multi-probe temperature verification system. If the Mastercycler fails using the second system it can be adjusted using the Eppendorf multi-probe temperature verification system (DOC ID [2919](#)). A performance check, in the form of a temperature verification, is then required before the Mastercycler can be returned into service.

Document ID	Revision	Approval	Date Published
7496	8	Supervising Criminalist - Biology	8/28/2018 9:53:16 AM

F.14 Insert a MultiMediaCard™ (MMC) into the control panel in the appropriate port. The control panel will beep, indicating it recognizes the card. Press "Export". A new screen will appear asking the user if they want to save the file as a PDF or a text only file. Make sure PDF is selected and press "OK". A new message will appear showing the export was successful. Press "OK".

NOTE: USB DRIVES CANNOT BE USED FOR EXPORTING DATA FILES FOR THE MASTER CYCLER EP'S. IT CAN ONLY BE USED FOR MASTER CYCLER PRO'S.

F.15 The certificate screen will reappear. Press "Exit". A message will appear to open the thermal cyclers. Press "OK". The control panel reverts to the original control panel screen.

F.16 Repeat the protocol for the remaining cyclers. Be sure to connect the unit to the appropriate thermal cycler for each run before continuing.

F.17 After verification of all cyclers, remove the MMC and place the run files in the appropriate location (K:\DIVISION\DNA\Thermal Cycler Temperature Verification\Single Probe TVS). Give the run files their own unique file with the date of the run. Rename the run files appropriately.

F.18 Print out all verification certificates and place them in the Temperature Checks binder. Disassemble the probe and return it to its case.

G. INTERPRETATION GUIDELINES

N/A

H. REFERENCES

H.1 Testo Instruction Manual, 2002.

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